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Additive Response of Biofertilizers on Seed Yield and Quality in Cowpea

R. Pavithra, S. Padmavathi, B. Sunil Kumar, S. Suganthi, P. Satheeshkumar, A. Kamaraj 10.18805/LR-4934

ABSTRACT

Background: Cowpea is a precious crop for farmers with limited resources. The crop's yield potential was constrained coupled with poor seed management methods, a lack of high yielding varieties and improper input use. Seed inoculation with biofertilizer is critical for increasing nutrient utilization efficiency and crop yield *via* nitrogen fixation and nodulations. With this in mind, a study was performed to appraise the influence of pre-sowing seed inoculation on initial seed quality, crop growth, seed yield, seed biochemical content and seed quality in cowpea, both in the laboratory and field.

Methods: Cowpea seeds were imposed with different bio-inoculants *viz*., *Rhizobium* @ 600 g/ha, *PSB* @ 600 g/ha, *VAM* @ 600 g/ha and *Pseudomonas* @ 10 g/kg as single treatment and in combined inoculations. The above treated seeds along with control were evaluated for seed quality under laboratory condition and crop growth and yield potential under field condition.

Result: *Rhizobium* inoculation (600 g/ha) and *phosphate solubilizing bacteria* inoculation (600 g/ha) were added to the seeds before they were sown. This led to better growth, yield and seed quality.

Key words: Biochemical content, Cowpea, PSB, *Rhizobium,* Seed quality, Seed yield.

INTRODUCTION

India seems to be the world's largest producer of pulses, accounting for 25.7 per cent of the world output, of which 27 per cent is for global consumption and 14 per cent is for imports. Cowpea is an excellent component of nutritional protein in semi-arid Africa and Asia and a precious crop for farmers with limited resources. The crop's yield potential was limited due to poor seed management procedures, an absence of high - yielding varieties and improper input use. It is grown primarily in arid and semi-arid regions of Punjab, Haryana, Delhi and West UP and in significant parts of Rajasthan, Karnataka, Kerala, Tamil Nadu, Maharashtra and Gujarat.

Seed is critical for agriculture's continued expansion, as it is the fundamental and core resource for multiplication. Seed quality has been revered as a sacred being and a critical ingredient in agriculture's and agricultural cultures' development. No agricultural procedure can develop a crop beyond the restrictions established by the seeds. Only through seeds can any advancement in crop enhancement be utilized and established in the land. Consequently, the agricultural industry requires and values high-quality seed production. Successful seed production requires various techniques to increase the productivity of high-quality seed, as seed production is influenced by seed, pre-harvest and post-harvest crop management techniques (Copeland and McDonald, 1985), cautioning managers to exercise caution during each stage of the seed production cycle.

The emergence of seedling in the field is a key and primary requirement for improved crop output. This is strongly reliant on the viability and vigour of the seeds used during sowing. To accomplish this goal, a holistic approach is required, including the usages of solid and established technologies, scientific management techniques for seed production and the provision of high-quality seeds. The seed's poor performance was an outcome of a reduction in vigour and viability. Seed quality may indeed vary between cultivars, including across and between seed lots. Adopting effective pre-sowing seed management procedures is among the enhanced seed production packages that increased production and yield.

Agents can be supplied "in the right amount, at the right place and at the right time" *via* seed treatment. There is a variety of pre-sowing seed treatments to gain higher agricultural production. Nevertheless, due to shortages of chemicals and their increased price, such methods are still not conceivable for farmers. Several of the pre-sowing seed treatments, such as seed pelleting, can boost the germination of seeds and vigour, particularly in unfavourable environmental conditions. Inoculation is the method of inoculating a seed with an effective growth promoting microorganism just before sowing. Inoculation aims to confirm

Department of Genetics and Plant Breeding, Faculty of Agriculture, Annamalai University, Annamalai Nagar-608 002, Chidambaram, Tamil Nadu, India.

Corresponding Author: A. Kamaraj, Department of Genetics and Plant Breeding, Faculty of Agriculture, Annamalai University, Annamalai Nagar-608 002, Chidambaram, Tamil Nadu, India.
Email: a.kamaraj@yahoo.co.in

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that the whole of the right type of microorganism is present in the soil to support a successful crop-organism symbiosis.

Seed inoculation of biofertilizer is critical for increasing nutrient usage efficiency and crop yield through nitrogen fixation and nodulations, especially in pulse crops (Raj and Raj, 2021). A bioinoculum constituted of competitive and effective *rhizobia* available to fix atmospheric nitrogen in conjunction with the activity of phosphorus solubilizing bacteria culminates in sustainable agriculture (Patel *et al*., 2014). Only living microbes are considered biofertilizers; they promote growth and reproduction, ensure sufficient nourishment to the host plants and maintain their healthy growth and function in their physiology (Khan *et al*., 2018). The purpose of this study was to determine the impact of pre-sowing seed treatment on crop growth, yield components and seed quality in cowpea cv. Vamban 3.

MATERIALS AND METHODS

A genetically and physically pure seeds of cowpea cv. Vamban 3 was procured from National Pulses Research Centre, Vamban, Pudukkottai which served as a basic material for the study. The experiments were carried out in 2019-2020 under Laboratory and field conditions in the Department of Genetics and Plant Breeding, Faculty of Agriculture, Annamalai University. The seeds were imposed with the pelleting treatment as given in the pre-sowing seed treatment details. For pelleting, the seeds are taken and smeared with an adhesive (10% Maida solution) followed by shaking gently to spread the adhesive materials evenly on each seed. Then, pelleting materials were added as per the treatment evenly over the seeds and continue shaking until the uniform coating was ensured. The pelleted seeds were dried back to the original moisture content. Both pelleted and unpelleted seeds (control) were used for evaluation of their quality (ISTA, 2019) and performance under field condition.

Pre-sowing seed treatments details

T_0 - Control
T_1 - Rhizobium @ 600 g/ha
T_2 - PSB @ 600 g/ha
T_3 - VAM @ 600 g/ha
T_4 - Pseudomonas fluorescens @ 10 g/kg
T_5 - Rhizobium @ 600 g/ha + PSB @ 600 g/ha
T_6 - Rhizobium @ 600 g/ha + VAM @ 600 g/ha
T_7 - Rhizobium @ 600 g/ha + Pseudomonas fluorescens @ 10 g/kg
T_8 - PSB @ 600 g/ha + Pseudomonas fluorescens @ 10 g kg
T_9 - VAM @ 600 g/ha + Pseudomonas fluorescens @ 10 g/kg
T_{10} - PSB @ 600 g/ha + VAM @ 600 g/ha

The above treated seeds along with control (unpelleted seeds) were sown in the sand medium under laboratory condition by adopting a completely randomized block design with three replications and observed for the seed quality parameters. The field trial was conducted with the above pelleted seeds along with control by adopting a randomized block design replicated thrice to evaluate the efficacy of pre-sowing seed treatment on crop growth and yield parameters during the year 2019. The resultant harvested seeds were assessed for their seed quality. The biochemical parameters *viz*., albumin, globulin, protein and NPK content in seeds also were recorded for the resultant seeds. The data from lab and field trials were analyzed statistically adopting the procedure described by Panse and Sukhatme (1985).

RESULTS AND DISCUSSION

In the present study, seeds subjected to various pre-sowing seed treatments are tested for their initial seed quality under laboratory and crop growth and yield of cowpea under field conditions. In laboratory analysis, the combined inoculation of *Rhizobium* and PSB treated seeds recorded higher values for the initial seed qualities *viz*. speed of germination (12.87), germination percentage (97), shoot length (26.70 cm), root length (20.68 cm), seedling fresh weight (9.86 cm), seedling dry weight (0.75 g), Vigour Index I (4579) and Vigour Index II (72) and the lower values were recorded in control (T_0) as represented in Fig 1.

The isolate's ability to synthesize seed germination hormones like gibberellins may have prompted the action of particular enzymes like α-amylase, which increase the availability of starch for absorption and hence promote early germination. It may also be due to improved mitochondrial enzyme activity, higher oxygen consumption and increased absorption of carbon nitrogen ratio. Inoculation of *Rhizobium* and PSB has brought this favourable result. The poor germination from untreated seed may be due to their failure to mobilize the resource from the seeds during initial period of germination but inoculated seeds made of the loss by using an improved synthesis of secondary metabolites and the presence of growth promoting substanceswhich migrates into the seeds have brought this positive effect. These resultsconform with the findings of Mandal (2018) in cowpea and Jaya *et al.* (2018) in peanut and soybean.

Rhizobium @ 600 g/ha + PSB @ 600 g/ha (T_5) recorded significantly higher values for root length, shoot length, seedling fresh and dry weight, seedling Vigour Index I and seedling Vigour Index II. There was 27.79%, 26.26%, 28.19% 14.67%, 31.51% and 18.06% increase in shoot length, root length, seedling fresh and dry weight, seedling Vigour Index I and II (Table 1) over the control respectively. Increment in seedling characters might be due to the process of repair mechanism and more synchronized germination which leads to early germination and early vigour with rapid rate of emergence because of which the seedling had reached autotrophic stage well in advance than T_0 and the production of longer seedlings. Amruta *et al.* (2016) in blackgram, Chauhan *et al.* (2016) and Kumar and Pandita (2016) in cowpea and Raja and Takankhar (2017) and Jaya *et al.* (2018) in soybean have found similar results.

In the current study, a field trial was conducted with seeds subjected to pre-sowing seed treatment using various bio-inoculants as mentioned above to assess the effect of bioinoculant on the growth and yield parameters of cowpea under field condition. Among the treatments, it was observed that combined inoculation of *Rhizobium* and PSB treated seeds (T_5) recorded higher values for the growth traits *viz.* field emergence (97%), plant height(67.42 cm), number of branches plant^{-1}(5.00), number of leaves plant^{-1} (67.73), leaf

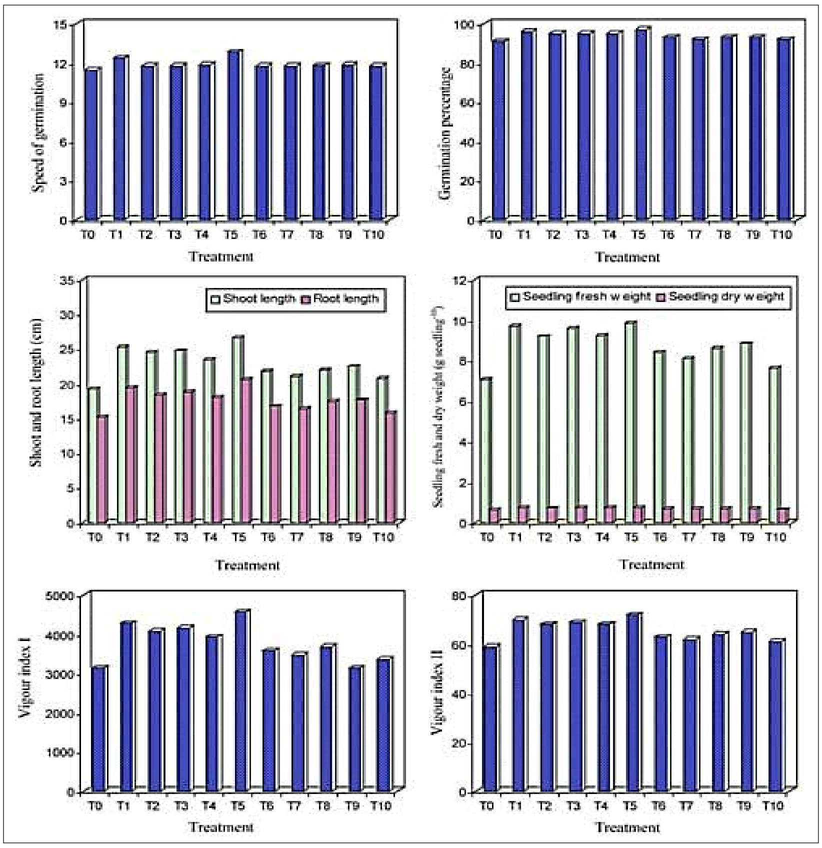

Fig 1: Effect of pre-sowing seed treatment on seedling quality characteristics of cowpea cv. Vamban 3.

Table 1: Effect of pre-sowing seed treatment on field emergence, plant height, number of leaves/plant, leaf length, leaf breadth and days to first flowering of cowpea cv. Vamban 3.

Treatment	Field emergence (%)	Plant height @ 30DAS (cm)	Number of leaves/plant	Leaf length (cm)	Leaf breadth (cm)	Days to first flowering
T_0	93 (77.12)	47.34	55.33	8.11	6.75	45.47
T_1	96 (79.05)	54.45	65.60	10.05	7.86	39.33
T_2	95 (77.59)	53.85	62.87	9.65	7.56	40.27
T_3	96 (78.00)	53.46	64.53	9.85	7.65	39.73
T_4	95 (77.12)	52.65	61.53	9.45	7.46	40.67
T_5	97 (79.51)	55.78	67.73	10.54	8.15	38.33
T_6	94 (76.24)	50.15	57.53	8.85	7.13	42.33
T_7	94 (75.86)	49.82	57.27	8.75	7.05	43.47
T_8	94 (76.24)	50.65	58.87	9.06	7.28	41.73
T_9	95 (75.10)	51.47	59.93	9.30	7.35	41.20
T_{10}	94 (75.47)	48.65	56.13	8.57	6.90	44.27
Mean	95 (77.03)	51.66	60.67	9.29	7.38	41.53
S.Ed	0.81	0.07	0.29	0.03	0.03	0.11
CD(P=0.05)	1.68	0.15	0.61	0.07	0.05	0.22

(Figures in parenthesis indicate arcsine transformed values).

length (10.54 cm), leaf breadth (8.15 cm), duration to first flowering (38.33 DAS) (Table 1) and duration to 50 per cent flowering (44.53 DAS) and yield attributing traits *viz.*, number of pods plant^{-1} (39.60), pod length (19.96 cm), number of seeds pod^{-1} (15.00), seed yield plant^{-1} (46.81 g) and 100 seed weight (14.92 g)(Fig 2) whereas the lower values for the above traits were registered in T_0 (control).

In the present study, seed treated with combined inoculation of *Rhizobium* and PSB(T_5) recorded the higher plant height, number of branches, number of leaves, leaf length and leaf breadth which was 12.24%, 20.00%, 18.31%, 23.06% and 17.18% increase over control respectively (Table 1). The minimum days to first flowering (38.33 DAS) (Table 1) and days to 50% flowering (44.53 DAS) (Fig. 2), were recorded in T_5 treated seeds which are 18.63% and 18.28% earlier than control respectively (Table 1). Rhizobium produces amino acids and growth promoters (auxin, cytokinin and GA) which fix atmospheric nitrogen, while PSB makes phosphorus available by solubilizing insoluble phosphates through the production of organic acids such as lactic acid and acetic acid during early plant growth of cowpea leading to improvement of the vegetative growth (crop canopy), the reduced number of days to first flowering and increased physiological activity of the plants resulted in earlier flowering and field emergence of T_5 plants (Mandal, 2018 in cowpea; Nadeem *et al.* (2017) in cowpea; Raja and Takankar (2017) in soybean and Singh *et al.* (2016) in blackgram).

In the present study, the number of pods plant^{-1}, pod length, number of seeds pod^{-1}, seed yield plant^{-1} and 100 seed weight registered 16.49%, 11.82%, 17.33%, 23.91% and 20.51% (Fig 2) higher than the untreated seeds respectively. Treatment T_5 increased the seed yield parameters might be due to increased availability of N and P in the root zone during the vegetative growth of crop through more atmospheric nitrogen fixation by rhizobium and solubilization of unavailable phosphates in the soil by PSB and which is not in case of biofertilizer inoculated alone or as single inoculation (Kalegore *et al.* (2018); Singh *et al.* (2018) in chickpea; Sibponkrung *et al.* (2020) in soyabean; Benjelloun *et al.* (2021) in chickpea). The combined inoculation of *Rhizobium* + PSB has involved in hormonal regulation, promoting pollen germination and pollen tube growth which in turn housed a maximum number of developed pods per plant (Raj *et al.* (2021) and Mandal (2018) in cowpea; Raklami *et al.* (2019) in Fababean; Dumsane *et al.*, 2020).

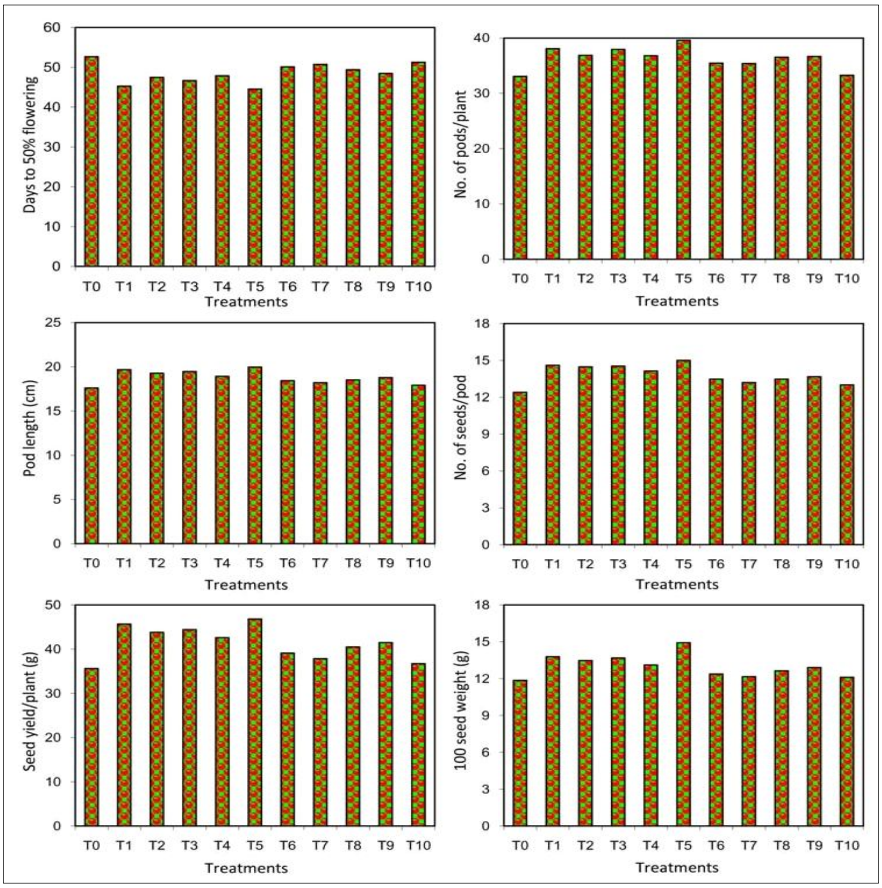

Fig 2: Effect of pre-sowing seed treatment on days to 1st flowering, days to 50% flowering, number of pods per plant, pod length, number of seeds /pod, seed yield/plant and 100 seed weight of cowpea cv. Vamban 3.

It was concluded that, an increase in the number of pods plant^{-1}, pod length, number of seeds pod^{-1}, seed yield plant^{-1} and 100 seed weight significantly increased the yield of cowpea.

In the present study, combined inoculation of rhizobium and phosphorous solubilizing bacteria inoculated seeds recorded higher values for the resultant seed qualities *viz*., speed of germination (12.97), germination percentage (97%), shoot length (16.74 cm), root length (20.73 cm), seedling fresh weight (9.85 cm), dry matter production (0.77 g), Vigour Index I (4579) and Vigour Index II (72) and lower values were recorded in control (Table 2). The superiority of the resultant seed from T_5 could be due to the presence of a higher number of metabolites/stored reserves (better translocation from source to sink) which helps in the resumption of embryonic growth during germination and also due to the accumulation of higher quantity of seed constituents (stored mRNA) and better DNA repair mechanism during germination and seedling emergence which in turns results in higher vigour.

The bio-chemical parameters *viz*., protein content, albumin content, globulin content and NPK content have significantly increased the resultant seed quality of cowpea (Fig 3). In the present study, T_5 (seed treatment with *Rhizobium* and PSB) recorded the higher values for albumin, globulin and protein content (4.30%, 0.254%, 22.73%) (Table 2) and NPK content (1.72%, 0.43%, 1.63%) (Fig 3) compared to control (T_0) respectively. The positive influence on nutrient content might be due to its impact on the carbon cycle in the plant *i.e*., higher CO_2 fixation and their efficient translocation toward developing seeds. The maximum uptake of these nutrients in this treatment may be owing to better root development as well as more nutrient availability by these microorganisms, resulting in better absorption and utilization of all plant nutrients leading to increased photo-assimilates, thus resulting in more nitrogen, phosphorus and potassium content in seed (Pardhi *et al*. (2022), Ramya *et al*. (2021) and Khan *et al*. (2017) in cowpea and Benjelloun *et al*. (2021) in chickpea.

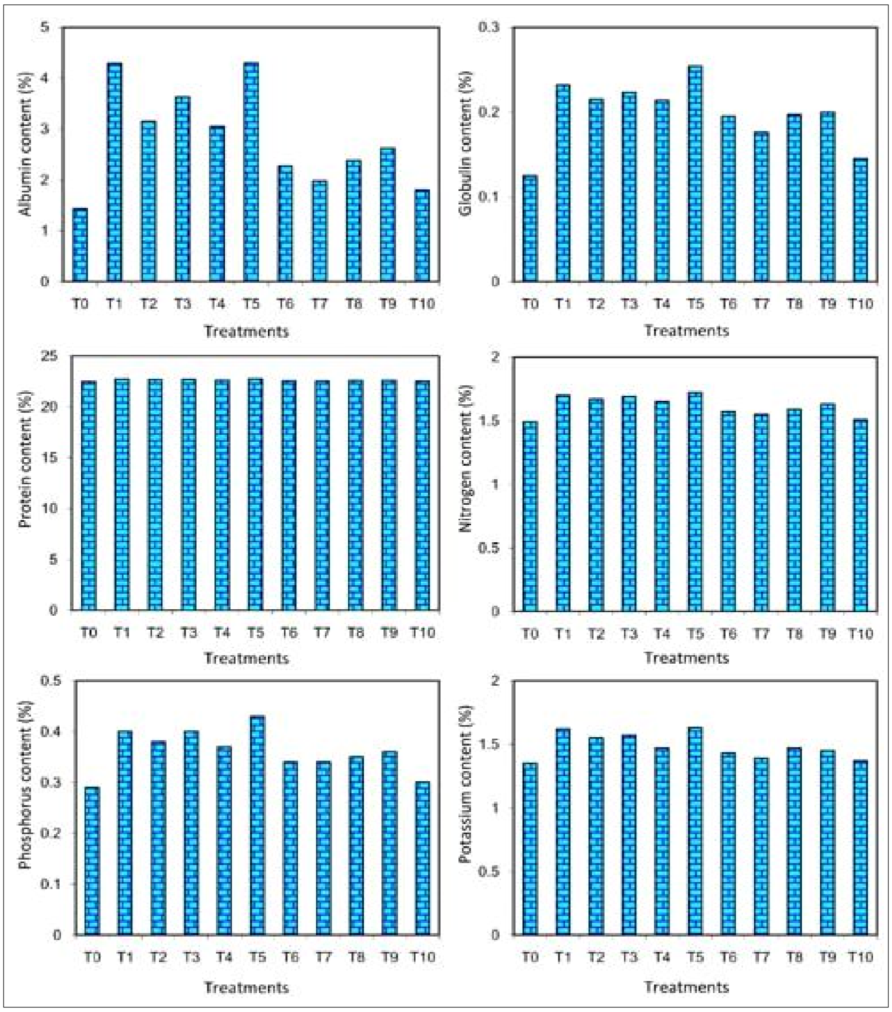

Fig 3: Effect of pre-sowing seed treatment on biochemical seed quality parameters *viz.,* Albumin content, Globulin content, Protein content and NPK content of harvested seeds of cowpea cv. Vamban 3.

Table 2: Effect of pre-sowing seed treatment on seedling quality of harvested seeds of cowpea cv. Vamban 3.

Treatment	Speed of germination	Germination %	Shoot length (cm)	Root length (cm)	Seedling fresh weight (g seedling^{-10})	Seedling dry weight (g seedling^{-10})	Vigour Index I	Vigour Index II
T_0	11.41	93 (74.29)	12.57	15.82	6.06	0.56	3136	59
T_1	12.87	96 (78.52)	16.23	19.17	8.41	0.76	4290	70
T_2	12.38	95 (77.54)	15.17	18.33	8.60	0.75	4087	68
T_3	12.59	95 (77.58)	15.84	18.86	8.47	0.75	4162	69
T_4	11.87	95 (77.54)	14.82	18.07	7.85	0.73	3935	68
T_5	12.97	97 (79.50)	16.74	20.73	9.85	0.77	4579	72
T_6	11.79	94 (76.24)	13.86	17.23	7.14	0.67	3590	63
T_7	11.47	94 (75.43)	13.45	17.06	7.94	0.66	3474	62
T_8	11.83	94 (76.27)	14.13	17.40	7.31	0.67	3682	64
T_9	11.84	95 (77.12)	14.45	17.82	7.57	0.72	3146	65
T_{10}	11.44	93 (75.14)	13.16	16.90	6.85	0.65	3369	61
Mean	12.04	95 (76.84)	14.58	17.94	7.82	0.70	3768	66
S.ED	0.01	0.39 (1.02)	0.08	0.24	0.19	0.02	281.02	1.35
CD (0.05)	0.03	0.85 (2.11)	0.17	0.49	0.39	0.03	582.81	2.79

CONCLUSION

Bioinoculum based on competitive and efficient rhizobia fixing atmospheric nitrogen coupled with P solubilizing bacteria could provide a feasible versatile and beneficial alternative to N and P chemical fertilizers for sustainable and environmentally friendly agriculture. From the present experiment, it was revealed that cowpea seeds inoculated with *Rhizobium* and phosphate solubilizing bacteria excelled in performance than other treatments.

Conflict of interest: None.

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